

**AMENDMENTS TO THE CLAIMS**

This listing of claims will replace all prior versions, and listings, of claims in the application.

**LISTING OF CLAIMS:**

1. – 12.       Cancelled.

13. (Currently Amended) A method for producing an evolved protein comprising:

- a)       generating a directed genetic modification in a gene of interest in an initial microorganism to yield a modified microorganism, wherein the production or consumption of a substrate is inhibited when the modified microorganism is grown on a defined medium, wherein the ability of the modified microorganism to grow is impaired;
- b)       culturing the modified microorganism obtained in step (a) on the defined medium allowing the modified microorganism to evolve a ~~compensatory~~ metabolic pathway to compensate for the impaired growth, wherein the defined medium can contain a co-substrate promoting the evolution; [[and]]
- c)       selecting an evolved microorganism from step (b) able to grow on the defined medium, wherein at least one protein has evolved in the metabolic pathway ~~compensating for the inhibition~~ allowing the modified microorganism to proliferate; and
- d)       isolating the evolved protein.

14.       (Original) The method as claimed in Claim 13, characterized in that the produced evolved protein is purified.

15. – 37.       Cancelled.

38.       (Previously Presented) The method of claim 13, wherein the gene of interest is a homologous gene or a heterologous gene.

39.       (Previously Presented) The method of claim 13, wherein the evolved protein is a homologous protein or a heterologous protein.

40. (Previously Presented) The method of claim 13, wherein the defined medium is substantially free of the substrate the production or consumption of which is inhibited in the modified microorganism.

41. (Previously Presented) The method of claim 13, wherein, in step (b) a co-substrate is added to the defined medium.

42. (Currently Amended) The method of claim 13, wherein, in step [(c)] (b)a co-substrate is added to the defined medium.

43. (Currently Amended) The method of claim 13, wherein the protein having evolved in the compensatory metabolic pathway is encoded by a gene being a homologous gene or a heterologous gene.

44. (Previously Presented) The method of claim 13, wherein the genetic modification comprises the directed mutation or deletion of a gene of interest or the directed modification of a promoter in the gene of interest.

45. (Previously Presented) The method of claim 13, wherein the genetic modification consists in the removal of most of the gene of interest.

46. (Previously Presented) The method of claim 13, wherein the gene of interest is replaced with a selection marker gene.

47. (Previously Presented) The method of claim 13, wherein the microorganism is selected among bacteria, yeasts and fungi.

48. (Previously Presented) The method of claim 13, wherein the microorganism is *Aspergillus sp.*, *Bacillus sp.*, *Brevibacterium sp.*, *Clostridium sp.*, *Corynebacterium sp.*,

*Escherichia sp.*, *Gluconobacter sp.*, *Pseudomonas sp.*, *Rhodococcus sp.*, *Saccharomyces sp.*,  
*Streptomyces sp.*, *Xanthomonas sp.*, or *Candida sp.*

49. (Currently Amended) The method of claim 13, wherein the microorganism is *E. coli*  
[[and]] or *C. glutamicum*.